

3874-129 US



2

## WHAT IS CLAIMED IS:

- 4 1. Composition for inducing activation of dendritic cells comprising a  
polynucleotide, viral vector, or polynucleotide derivative thereof and at  
6 least one polyoxyethylene-polyoxypropylene block copolymer.
- 8 2. The composition of claim 1 further comprising a polycation.
- 10 3. The composition of claim 2 wherein the polycation is a polyamine polymer.
- 12 4. The composition of claim 2 wherein the polycation is an oligoamine or an  
oligoamine conjugate.
- 14 5. The composition of claim 1 wherein there is a mixture of block copolymers.
- 16 6. The composition of claim 5 wherein the block copolymers comprise a  
mixture wherein at least one block copolymer with oxyethylene content of  
18 50% or less, and at least one block copolymer with oxyethylene content of  
20 50% or more.
- 22 7. The composition of claim 5 6 wherein the block copolymers comprise a  
mixture a first block copolymer component having an oxyethylene content  
24 of 50% or less, and a second block copolymer component having an  
oxyethylene content of 50% or more wherein the weight ratio of said  
26 second block copolymer to said first block copolymer is at least 2:1.
- 28 8. The composition of claim 6 wherein the block copolymers comprise a  
mixture a first block copolymer component having an oxyethylene content

3874-129 US

2 of 50% or less, and a second block copolymer component having an  
oxyethylene content of 50% or more wherein the weight ratio of said  
4 second block copolymer to said first block copolymer is at least 5:1.

6 9. The composition of claim 5 wherein the copolymers comprise a mixture  
wherein at least one block copolymer has an oxyethylene content of 70%  
8 or more and at least one block copolymer has an oxyethylene content of  
50% or less.

10

10. The composition of claim 5 wherein N, according to the following  
12 expression, is from about 0.2 to about 9.0 and preferably from about 0.25  
to about 1.5:

14

$$N = 1.32 \cdot \left[ \frac{H_1 \cdot m_1}{(L_1) \cdot (m_1 + m_2)} + \frac{H_2 \cdot m_2}{(L_2) \cdot (m_1 + m_2)} \right]$$

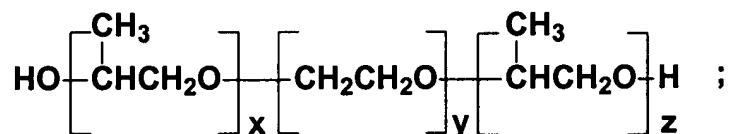
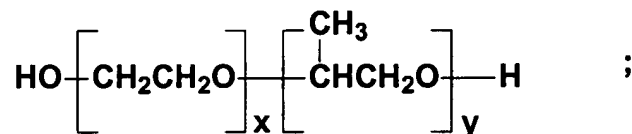
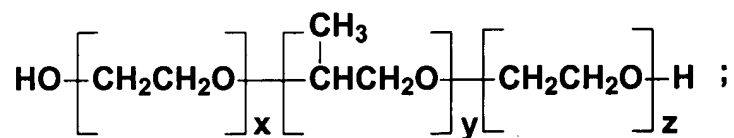
16 in which  $H_1$  and  $H_2$  are the number of oxypropylene units in the  
first and second block copolymers, respectively;  $L_1$  is the number of  
18 oxyethylene units in the first block copolymer;  $L_2$  is the number of  
oxyethylene units in the second block copolymer;  $m_1$  is the weight  
20 proportion in the first block-copolymer; and  $m_2$  is the weight proportion  
in the second block copolymer.

22 11. The composition of claim 5 wherein the mixture comprises the block  
copolymer PLURONIC® F127.

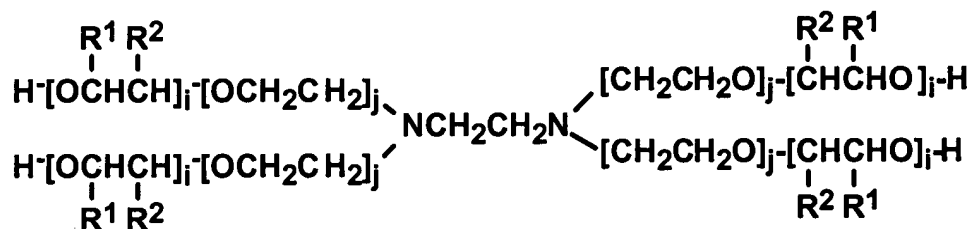
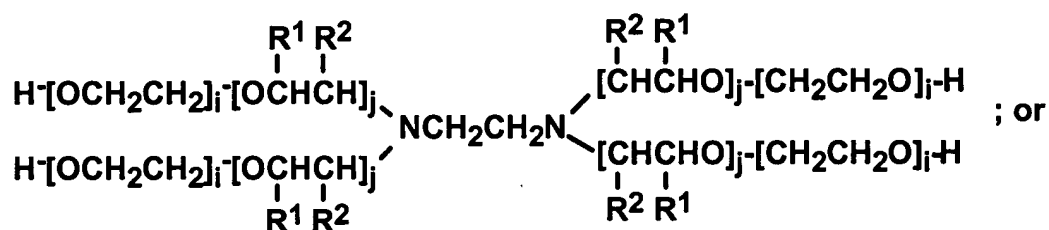
24

12. The composition of claim 1, wherein at least one of the block copolymers  
26 has the formula:

3874-129 US



2

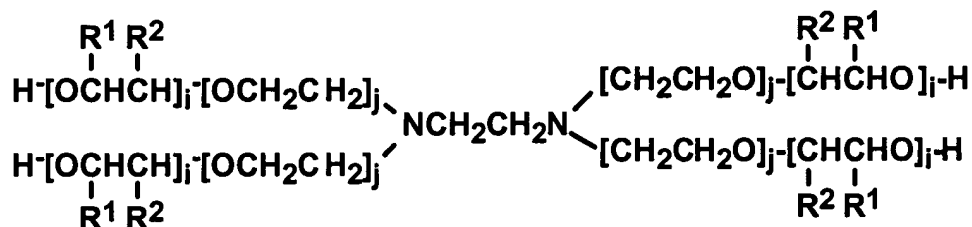
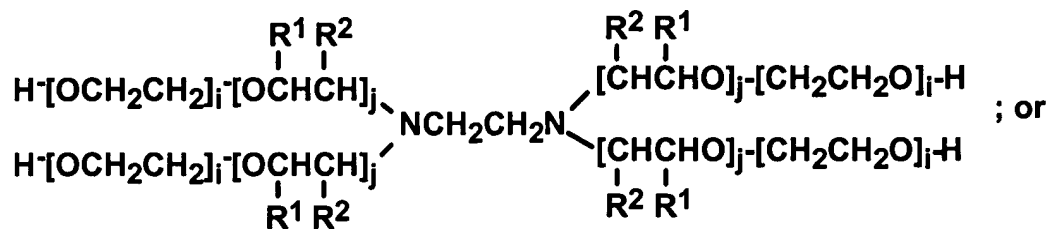


4

6 in which  $x$ ,  $y$ ,  $z$ ,  $i$ , and  $j$  have values from about 2 to about 400, and  
 6 wherein for each  $\text{R}^1$ ,  $\text{R}^2$  pair, one is hydrogen and the other is a methyl group.

8 13. The composition of claim 1 wherein at least one of the block copolymers  
 has the formula:

3874-129 US



in which  $x$ ,  $y$ ,  $z$ ,  $i$ , and  $j$  have values from about 2 to about 400,  
and for each  $\text{R}^1$ ,  $\text{R}^2$  pair, one is hydrogen and the other is a methyl  
group.

14. The composition of claim 1 wherein the block copolymer comprises at least  
PLURONIC F127 and L61.

15. The composition of claim 14 wherein the ratio of PLURONIC F127:L61 is  
8:1.

16. The composition of claim 14 wherein PLURONIC F127 is in the amount of  
about 2%w/v and PLURONIC L61 is in the amount of about 0.025% w/v.

17. The composition of claim 1 wherein said block copolymer is present in  
amounts insufficient for gel formation.

18. A composition for inducing the activation of dendritic cells comprising a  
polynucleotide or derivative thereof and at least one polyoxyethylene-

3874-129 US

2 polyoxypropylene block copolymer, wherein the block copolymer is present  
at a concentration below about 15% wt/vol.

4

19. The composition of claim 18 wherein the block copolymer concentration is  
6 below about 10%.

8 20. The composition of claim 18 wherein the block copolymer concentration is  
below about 5%.

10

21. A composition for inducing the activation of dendritic cells comprising a  
12 polynucleotide or derivative thereof and at least one polyoxyethylene-  
polyoxypropylene block copolymer, wherein the composition forms a  
14 molecular solution or colloidal dispersion.

16 22. The composition of claim 21 wherein the colloidal dispersion is a  
suspension, emulsion, microemulsion, micelle, polymer complex, or other  
18 type of molecular aggregate.

20 23. The composition of claim 21 wherein the colloidal dispersion comprises  
molecular species that are less than about 300 nm.

22

24. The composition of claim 21 wherein the colloidal dispersion comprises  
24 molecular species that are less than about 100 nm.

26 25. The composition of claim 21 wherein the colloidal dispersion comprises  
molecular species that are less than about 50 nm.

28

26. The composition of claim 1 wherein the polynucleotide is RNA, DNA,  
30 plasmid DNA, virus, or viral vector.

3874-129 US

- 2 27. The composition of claim 1 wherein the polynucleotide encodes a secreted  
or non-secreted protein, vaccine or antigen.
- 4
- 6 28. The composition of claim 1 comprising a gene expressing a secreted or  
non-secreted protein, vaccine or antigen and at least one gene expressing  
an adjuvant molecule operable to activate antigen presenting cells and  
8 induce immune response for enhanced antigen presentation.
- 10 29. A method of inducing activation of dendritic cells comprising administering  
a composition comprising a polynucleotide or derivative thereof and at  
12 least one polyoxyethylene-polyoxypropylene block copolymer.
- 14 30. The method of claim 30 wherein the block copolymers comprise at least  
PLURONIC F127 and L61.
- 16
- 18 31. The method of claim 30 wherein the block copolymer is present in amounts  
insufficient for gel formation.
- 20 32. A method of inducing activation of dendritic cells comprising administering a  
composition comprising a polynucleotide or derivative thereof and at least  
22 one polyoxyethylene-polyoxypropylene block copolymer, wherein the  
composition forms a molecular solution or colloidal dispersion.
- 24
- 26 33. The method of claim 32 wherein the block copolymers are PLURONIC  
F127 and L61.
- 28 34. A method of increasing the immune response of an animal comprising  
administering the composition according to claim 1.
- 30

3874-129 US

2 35.The method of claim 34 wherein the block copolymers comprise at least  
PLURONIC F127 and L61.

4  
36.The method of claim 34 wherein the composition is administered orally,  
6 topically, rectally, vaginally, parenterally, intramuscularly, intradermally,  
subcutaneously, intraparietally, or intravenously.

8  
37.A method of increasing the immune response of an animal comprising  
10 intramuscularly administering the composition according to claim 1.

12 38.The method of claim 37 wherein the block copolymers comprise at least  
PLURONIC F127 and L61.

14 39.The method of claim 37 wherein said composition is administered to at  
least one of smooth, skeletal, and cardiac muscles.

16  
40.A method of increasing the immune response of an animal comprising  
18 intradermally administering the composition according to claim 1.

20 41.A composition for inducing the activation of dendritic cells comprising at least  
one polyoxyethylene-polyoxypropylene block copolymer.

22  
42.The composition of claim 41 further comprising a polycation.

24  
43.The composition of claim 42 wherein the polycation is a polyamine polymer.

26  
44.The composition of claim 41 wherein the polycation is an oligoamine or an  
28 oligoamine conjugate.

30 45.The composition of claim 41 wherein there is a mixture of block  
copolymers.

3874-129 US

2

4

6

46. The composition of claim 45 wherein the block copolymers comprise a mixture wherein at least one block copolymer with oxyethylene content of 50% or less, and at least one block copolymer with oxyethylene content of 50% or more.

8

10

47. The composition of claim 46 wherein the ratio by weight of the block copolymer with oxyethylene content of 50% or less to the block copolymer with oxyethylene content of 50% or more is 1:2.

12

14

48. The composition of claim 46 wherein the ratio by weight of the block copolymer with oxyethylene content of 50% or less to the block copolymer with oxyethylene content of 50% or more is 1:5.

16

18

20

49. The composition of claim 45 wherein the copolymers comprise a mixture wherein at least one block copolymer with oxyethylene content of 70% or more and at least one block copolymer with oxyethylene content of 50% or less.

22

50. The composition of claim 45 wherein N, according to the following expression, is from about 0.2 to about 9.0 and preferably from about 0.25 to about 1.5:

24

$$N = 1.32 \cdot \left[ \frac{H_1 \cdot m_1}{(L_1) \cdot (m_1 + m_2)} + \frac{H_2 \cdot m_2}{(L_2) \cdot (m_1 + m_2)} \right]$$

26

in which  $H_1$  and  $H_2$  are the number of oxypropylene units in the first and second block copolymers, respectively;  $L_1$  is the number of oxyethylene units in the first block copolymer;  $L_2$  is the number of oxyethylene units in



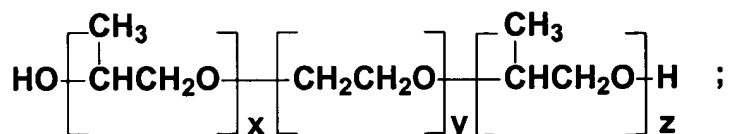
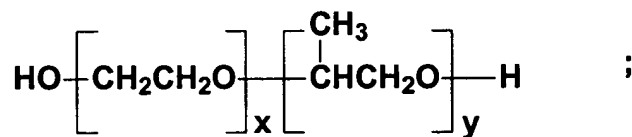
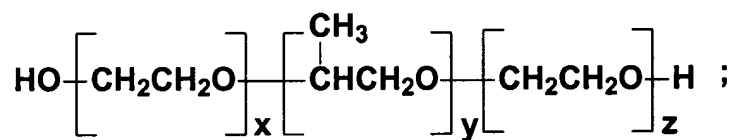
3874-129 US

2 the second block copolymer;  $m_1$  is the weight proportion in the first block-  
 4 copolymer; and  $m_2$  is the weight proportion in the second block copolymer.

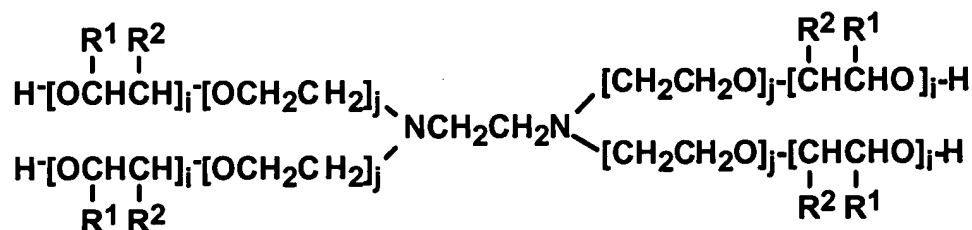
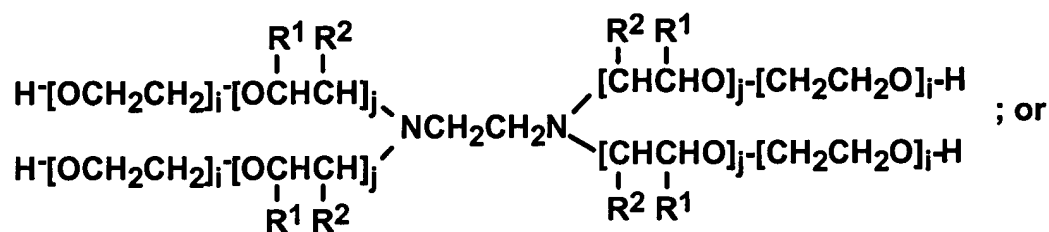
4

51. The composition of claim 45 wherein the mixture comprises the block  
 6 copolymer PLURONIC® F127.

8 52. The composition of claim 41, wherein at least one of the block copolymers  
 has the formula:

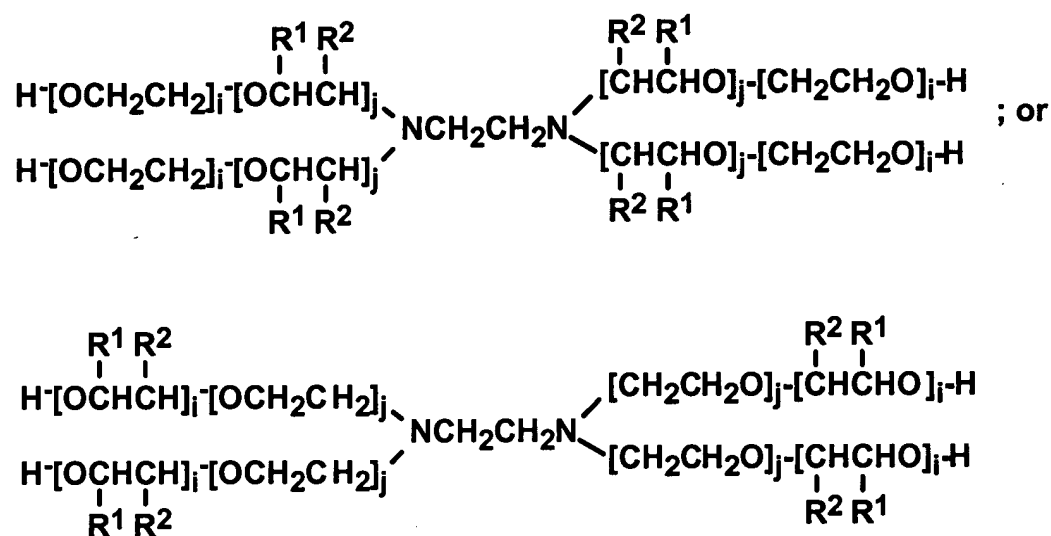


10



2 in which  $x$ ,  $y$ ,  $z$ ,  $i$ , and  $j$  have values from about 2 to about 400, and  
wherein for each  $R^1$ ,  $R^2$  pair, one is hydrogen and the other is a methyl  
4 group.

8



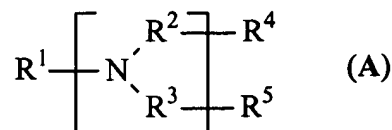
14

16

3874-129 US

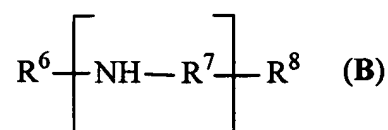
- 2 56. A composition for inducing the activation of dendritic cells comprising at  
least one polyoxyethylene-polyoxypropylene block copolymer, wherein the  
4 composition forms a molecular solution or colloidal dispersion.
- 6 57. The composition of claim 56 wherein the colloidal dispersion is a  
suspension, emulsion, microemulsion, micelle, polymer complex, or other  
8 type of molecular aggregate.
- 10 58. The composition of claim 56 wherein the colloidal dispersion comprises  
molecular species that are less than about 300 nm.
- 12 59. The composition of claim 56 wherein the colloidal dispersion comprises  
14 molecular species that are less than about 100 nm.
- 16 60. The composition of claim 56 wherein the colloidal dispersion comprises  
molecular species that are less than about 50 nm.
- 18 61. A composition for inducing activation of dendritic cells comprising a  
20 polynucleotide or derivative thereof and at least one polycationic polymer  
having a plurality of cationic repeating units.
- 22 62. A polynucleotide composition according to Claim 61 wherein said  
24 polycationic polymer is a cationic homopolymer, copolymer or block  
copolymer comprising one or more of the following fragments:
- 26 (a) at least one aminoalkylene monomer selected from a group  
consisting of:
- 28 (i) a tertiary amine monomer of the formula

3874-129 US



and,

(ii) a secondary amine monomer of the formula



in which:

each of  $R^1$ ,  $R^4$ ,  $R^5$ ,  $R^6$  and  $R^8$  taken independently of each other is hydrogen, alkyl of 2 to 8 carbon atoms, another A monomer, or another B monomer;

each of  $R^2$ ,  $R^3$  and  $R^7$ , taken independently of each other, is a straight or branched alkanediyl of the formula  $-(C_zH_{2z})-$  wherein  $z$  has a value of from 2 to 8;

(b) cationic amino acids;

(c)  $(-OPO(NH-R^9-NR^{10}R^{11}R^{12})O-R^8-)$  in which  $R^9$  is a stright chain aliphatic group of from 1-12 carbon atoms and  $R^8$  is  $-(CH_2)_n-CH(R^{13})-$  where  $n$  is an integer from 0 to 5,  $R^{10}$ ,  $R^{11}$  and  $R^{12}$  are independently hydrogen or alkyl of 1 to 4 carbon atoms and  $R^{13}$  is a hydrogen, cycloalkyl having 3-8 carbon atoms. or alkyl of 1-2 carbon atoms; and

(d) vinylpyridine or a derivative thereof.

63. A composition according to claim 62 comprising a polynucleotide and a polymer of a plurality of segments, wherein the polymers comprise at least one polycationic segment which is cationic homopolymer, copolymer, or block copolymer, or quaternary salt thereof; and either,

3874-129 US

2 (a) at least one straight or branched chain polyether segment of from 5  
to about 400 monomer units which polyether segment is:

4 (i) a homopolymer of at least one alkyleneoxy monomer  $-OC_nH_{2n}-$   
in which n has a value from 2 to 3; or,

6 (ii) a copolymer or block copolymer of said first alkyleneoxy  
monomer and a second different alkyleneoxy monomer  
8  $-OC_mH_{2m}-$ , in which n has a value from 2 to 3 and m has a value  
from 2 to 4 or,

10 (b) a homopolymer or copolymer of at least one monomer from a group  
consisting of acrylamide, glycerol, vinyl alcohol, vinyl pyrrolidine,  
12 vinylpyridine-N-oxide, oxazoline, morpholine acrylamide, and derivatives  
thereof.

14

64. A composition according to claim 63 wherein said first alkyleneoxy  
16 monomer is ethyleneoxy ( $-OCH_2CH_2-$ ) and said second alkyleneoxy  
monomer is propyleneoxy ( $-OCH(CH_3)CH_2-$ ).

18

65. A composition of claim 63 wherein the polycationic polymer, at  
20 physiological pH, comprise at least six cationic groups.

22 66. A polynucleotide composition according to claim 63 wherein the  
polycationic polymer, at physiological pH, contain a plurality of cationic  
24 groups separated by about 3Å to about 12Å.

26 67. A polynucleotide composition according to claim 63 wherein each of  
said polyether segments has from about 5 to about 80 monomeric units  
28 and said polycationic segment is a homopolymer, copolymer or block  
copolymer of from 2 to about 180 of the same or different monomeric  
units of the formula  $-NH-R^o-$  in which  $R^o$  is a straight chain aliphatic  
30 group of 2 to 6 carbon atoms which may be optionally substituted.

2

4

6

8

10

12

14

16

18

20

22

24

26

28

30

75. The method of claim 72 wherein the composition is administered orally, topically, rectally, vaginally, parenterally, intramuscularly, intradermally, subcutaneously, intraperitoneally, or intravenously.

76. The method of claim 72 wherein said composition is administered to at least one of smooth, skeletal, and cardiac muscles.

4

77. A method of improving the immune response of an animal comprising intradermally administering the composition according to claim 34.

8